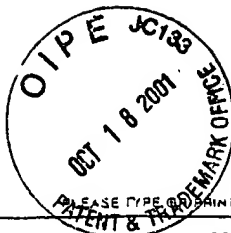


## ANIMAL USAGE FORM



AUF 1413

## 1. PRINCIPAL INVESTIGATOR

WAYNE A. BORDER, M.D.

OFFICE PHONE

226

HOME/EMERGENCY PHONE

(714) 770-4602

## 2. OTHER INVESTIGATOR

LUCIA LANGUINO, PH.D.

230

539-0609

## 3. SENIOR TECHNICIAN

## 4. PROJECT TITLE

ANTI-HUMAN TGF $\beta$  CYCLIZED PEPTIDE

## 5. GRANT NUMBER, IF ANY

250200

NEW

X

RENEWAL

PILOT

PROJECT NUMBER

## 6. START DATE

END DATE

MICE

RATS

RABBITS

GPI

OTHER (SPECIFY)

QUANTITY:

2

## 7. PROJECT GOAL (SEE INSTRUCTIONS)

To produce quantities of anti-human TGF $\beta$  cyclized peptide for use in kidney disease research.

## 8. RATIONALE (SEE INSTRUCTIONS)

Rabbits produce high quality antiserum which can be used for identification of human TGF $\beta$  in tissue samples and in vitro assays to study progression of kidney injury.

## 9. DESCRIBE USE OF ANIMALS (SEE INSTRUCTIONS)

All injections/bleedings to be performed by animal care facility personnel.

1. Pre-bleeding 20 ml from ear vein.
2. Inject 500  $\mu$ g TGF $\beta$  cyclized purified peptide (0.5 ml antigen in PBS + 0.5 ml FCA) subcutaneously in 2 sites.
3. After one month, boost with 125  $\mu$ g antigen (0.25 ml antigen in PBS + 0.25 ml incomplete adjuvant) subcutaneously, 2 sites.
4. After 10 days, bleed 50 ml from alternating ear veins 3 times.
5. Repeat steps 3-4 at 4-6 week intervals.

FOR ANIMAL RESEARCH COMMITTEE  
 REVIEW AND APPROVAL REQUIRED  
 ALL ANIMAL RESEARCH MUST BE  
 APPROVED BY THE ANIMAL CARE  
 AND USE COMMITTEE (ACUC) BEFORE  
 BEGINNING RESEARCH IN THIS  
 RESEARCH FACILITY

## 10. PAIN LEVEL

A

B

C

(IF B OR C READ INSTRUCTIONS. PROVIDE DESCRIPTION OR JUSTIFICATION HERE)

CONFIDENTIAL

## 11. EUTHANASIA (SEE INSTRUCTIONS)

DURING PROJECT

Y

METHOD  
OR  
TECHNIQUE

CO.

Y

CERV. DISLOC.

OTHER (SPECIFY)

RETAIN CARCASSES?

YES

END OF PROJECT

Y

O.D.

Y

FOR R

NO

## 12. SIGNATURES

PI

WABorder

DATE

AP

MGR

DATE

U2 05334

DATE

# ANIMAL PROCEDURE REQUEST

(For procedures to be performed by Animal Facility personnel)  
Return this form to the Animal Facility Office.

LJCRF

TODAY'S DATE: \_\_\_\_\_

Principal Investigator E. ROSLAHT

LUCIA R. LARGUINHO

Investigator/Technician

Phone

230

Lab No.

Phone

Lab No.

This procedure relates to the project outlined on Animal Usage Form No. \_\_\_\_\_

Date or Dates on which this procedure is required (please allow 7 days for scheduling):

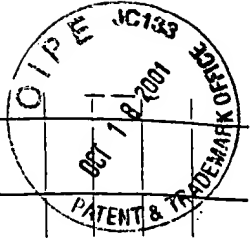
- 1 12/13/88 50 ml (If more than 8 procedures are required, attach a second form)
- 2 12/16/88 "
- 3 12/21/88 Estm 100 ml
- 4 \_\_\_\_\_

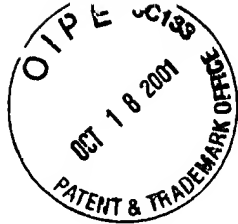
Describe the procedure(s) required:

Vol. of blood to be collected each procedure:

ml.

Injection 6 subcut 2 PC  
2 TGFβ Luciferase  
2 " " cyclic





Injection of 2 rabbits with Linear 78-103 peptide  
no 86-87 from TGF $\beta$ .

2 " with lyophilized 78-103 " .  
no 88-89

Procedure: 2 mg / rabbit of each peptide.

- 2 mg peptide were dissolved in 250  $\mu$ l DAW
- 0.5 mg Methylated BSA were ~~added to the solution to couple the peptide~~  
~~added to the solution to couple the peptide~~  
(Sigma A1003 : M<sub>2</sub>BSA)
- vortex 30''
- Added 250  $\mu$ l Freund's adjuvant complete
- mixed 1h with homogenizer.
- 0.5 ml solution was injected in each rabbit

Note : Both peptides were difficult to dissolve.  
" " were not HPLC purified



Injection of 2 rabbits linear TGF $\beta$  peptide

Cyclic

PG peptide

Procedure

except Freund's incomplete adjuvant was used

For notes:

KLH-peptide was already coupled

I only mixed to F.i. 20% in 1h 4.5

inject 0.5ml

Rabbit

n° 1284  
1285

for PG

ALF 1385